AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application.

Listing of Claims

Claims 1-33 (canceled).

Claim 34 (currently amended): An amplification reaction mixture for the quantitation of a target viral RNA ~~segment~~ sequence in a biological sample, said reaction mixture comprising:

said target viral RNA sequence;

a predetermined initial amount of ~~a control sequence for quantitation of a target viral RNA, wherein said control sequence and its complementary sequence bind the same primers as are bound by said target viral RNA segment and its complementary sequence~~ at least one reference RNA sequence selected from the group consisting of (i) a reference RNA sequence which does not include the target viral RNA sequence, (ii) a reference RNA sequence which includes substantially more nucleotides than the target viral RNA sequence, (iii) a reference RNA sequence which includes at least about 20 nucleotides less than the target viral RNA sequence and (iv) a reference RNA sequence which comprises target viral RNA sequence and non-target viral RNA sequence; and

an oligonucleotide primer pair ~~wherein said primer pair can serve to amplify said control sequence and said target viral RNA~~ for each of the target viral RNA sequence and the reference RNA sequence to be amplified, wherein following amplification ~~said control~~ amplified reference RNA sequence and amplified target ~~segments~~ viral RNA sequence are distinguishable by size or by probes.

Claim 35 (currently amended): A reverse transcription reaction mixture for reverse transcribing a target viral ~~mRNA~~ RNA sequence suspected of being present in a biological sample, said reaction mixture comprising:

a target viral RNA sequence;

a predetermined initial amount of ~~a control sequence cRNA, a target viral RNA,~~ at least one reference RNA sequence selected from the group consisting of (i) a reference RNA sequence which does not include target viral RNA sequence, (ii) a reference RNA sequence which includes substantially more nucleotides than target viral RNA sequence, (iii) a reference RNA sequence which includes at least about 20 nucleotides less than target viral RNA sequence and (iv) a reference RNA sequence which comprises target viral RNA sequence and non-target viral RNA sequence; and

~~a target-specific~~ an oligonucleotide primer pair for each of the target viral RNA sequence and the reference RNA sequence to be amplified for initiating cDNA synthesis to provide a target viral cDNA and a reference sequence cDNA, ~~wherein said primer can serve to initiate reverse transcription of a nucleic acid segment contained within said control sequence cRNA together with a segment contained within the particular target viral RNA, and wherein said control sequence is further distinguished by having a hybridization site identical in sequence to a hybridization site in said target viral RNA,~~ whereby following reverse transcription the resulting target viral and ~~control~~ reference sequence cDNAs can serve as templates for amplification for providing ~~control~~ amplified reference RNA sequence and amplified target ~~amplified~~ viral RNA ~~segments~~ sequence which are distinguishable by size or by probes.

Claims 36-37 (canceled).

Claim 38 (currently amended): The mixture of claim 34, wherein the ~~control sequence is a maxigene~~ reference RNA sequence comprises target viral RNA sequence and non-target viral RNA sequence.

Claim 39 (currently amended): The mixture of claim 35, wherein the ~~control sequence is a maxigene~~ reference RNA sequence comprises target viral RNA sequence and non-target viral RNA sequence.

Claims 40-41 (canceled).

Claim 42 (currently amended): The mixture of claim 34, wherein the target viral RNA sequence is contained within a nucleic acid sequence which encodes a protein associated with HIV or HCMV.

Claim 43 (currently amended): The mixture of claim 35, wherein the target viral RNA sequence is contained within a nucleic acid sequence which encodes a protein associated with HIV or HCMV.

Claim 44 (currently amended): A kit for the quantitation of a target viral RNA ~~segment~~ sequence in a biological sample comprising individual containers which provide:

a predetermined initial amount of ~~a control sequence for quantitation of a target viral RNA wherein said control sequence binds the same primers as are bound by said target viral RNA segment and its complementary sequence~~ at least one reference RNA sequence selected from the group consisting of (i) a reference RNA sequence which does not include the target viral RNA sequence, (ii) a reference RNA sequence which includes substantially more nucleotides than the target viral RNA sequence, (iii) a reference RNA sequence which includes at least about 20 nucleotides less than the target viral RNA sequence and (iv) a reference RNA sequence which comprises target viral RNA sequence and non-target viral RNA sequence; and

an oligonucleotide primer pair ~~wherein said primer pair can serve to amplify said control sequence and said target viral RNA~~ for each of the target viral RNA sequence and the reference RNA sequence to be amplified,

wherein following amplification ~~said control~~ amplified reference RNA sequence and amplified target ~~amplified~~ viral RNA ~~segments~~ sequence are distinguishable by size or by probes ~~use of an internal oligonucleotide probe, and wherein the target viral RNA is contained within a nucleic acid sequence which encodes a protein associated with HIV or HCMV~~.

Claims 45-49 (canceled).

Claim 50 (new): A process for quantitation of a target viral RNA sequence in a peripheral blood cell sample which comprises:

(i) selecting the target viral RNA sequence;

(ii) simultaneously subjecting

(a) the sample and

(b) a predetermined quantity of at least one reference RNA sequence, wherein the reference RNA sequence is selected from the group consisting of (i) a reference RNA sequence that does not include the target viral RNA sequence and (ii) a reference RNA sequence that has substantially more nucleotides than the target viral RNA sequence

to polymerase chain reaction amplification under conditions appropriate to simultaneously amplify the target viral RNA sequence if present in the sample and the reference RNA sequence to produce amplification products;

(iii) denaturing the amplification products produced in step (ii);

(iv) subjecting the denatured amplification products of step (iii) to hybridization conditions separately and sequentially with a labeled probe homologous to the target viral RNA sequence and

a labeled probe homologous to the reference RNA sequence and detecting the presence or absence of the target viral RNA sequence and the presence of the reference RNA sequence in the denatured amplification products of step (iii) by Southern blot hybridization with the labeled probes,

wherein one of the probes is removed from any sequence with which it is hybridized prior to the separate and sequential subjection of the denatured amplification products of step (iii) to hybridization with the other of the probes;

(v) determining whether the amplified target viral RNA sequence and the amplified reference RNA sequence hybridized with the probes homologous therewith; and

(vi) determining the relative quantitation of the target viral RNA sequence by comparison of the amount of signal obtained from the hybridized amplified target viral RNA sequence with the amount of signal obtained from the hybridized amplified reference RNA sequence.

Claim 51 (new): The process of claim 50, wherein the target viral RNA sequence is a human immunodeficiency virus (HIV) RNA sequence or a human cytomegalovirus (HCMV) RNA sequence.

Claim 52 (new): The process of claim 50, wherein a primer utilized in the polymerase chain reaction amplification includes a T-7 RNA polymerase binding sequence.

Claim 53 (new): A process for quantitation of a target viral RNA sequence in a sample which comprises:

(i) selecting the target viral RNA sequence;

(ii) simultaneously subjecting

(a) the sample and

(b) a predetermined quantity of at least one reference RNA sequence selected from the group consisting of (i) a reference RNA sequence which does not include the target viral

RNA sequence, (ii) a reference RNA sequence which includes the target viral RNA sequence and has substantially more nucleotides than the target viral RNA sequence, (iii) a reference RNA sequence which includes the target viral RNA sequence with at least about 20 nucleotides less than the target viral RNA sequence and (iv) a reference RNA sequence which comprises target viral RNA sequence and non-target viral RNA sequence

to polymerase chain reaction amplification under conditions appropriate to simultaneously amplify the target viral RNA sequence if present in the sample and the reference RNA sequence to produce amplification products;

(iii) denaturing the amplification products;

(iv) subjecting the denatured amplification products to hybridization conditions separately and sequentially with a labeled probe homologous to the target viral RNA sequence and a labeled probe homologous to the reference RNA sequence and detecting the presence or absence of the amplified target viral RNA sequence and the presence of the amplified reference RNA sequence in the denatured amplification products by Southern blot hybridization with the labeled probes,

wherein one of the probes is removed from any sequence with which it is hybridized prior to the separate and sequential subjection of the denatured amplification products of step (iii) to hybridization with the other of the probes;

(v) determining whether the amplified target viral RNA sequence and the amplified reference RNA sequence hybridized with the probes homologous therewith;

(vi) determining the relative amount of the target viral RNA sequence by comparing the amount of signal obtained from the hybridized amplified target viral RNA sequence and the amount of signal obtained from the hybridized amplified reference RNA sequence; and

(vii) determining the initial amount of target viral RNA sequence present in the sample by comparing the result of the determination in step (vi) to the predetermined quantity of the reference RNA sequence.

Claim 54 (new): The process of claim 53, wherein the target viral RNA sequence is a human immunodeficiency virus (HIV) RNA sequence or a human cytomegalovirus (HCMV) RNA sequence.

Claim 55 (new): The process of claim 53, wherein a primer utilized in the polymerase chain reaction amplification includes a T-7 RNA polymerase binding sequence.

Claim 56 (new): A process for quantitation of a target viral RNA sequence in a sample which comprises:

(i) selecting the target viral RNA sequence;

(ii) simultaneously subjecting

(a) the sample and

(b) a predetermined quantity of at least one reference RNA sequence selected from the group consisting of (i) a reference RNA sequence which does not include the target viral RNA sequence, (ii) a reference RNA sequence which includes the target viral RNA sequence and has substantially more nucleotides than the target viral RNA sequence, (iii) a reference RNA sequence which includes the target viral RNA sequence with at least about 20 nucleotides less than the target viral RNA sequence and (iv) a reference RNA sequence which comprises target viral RNA sequence and non-target viral RNA sequence

to polymerase chain reaction amplification under conditions appropriate to simultaneously amplify the target viral RNA sequence if present in the sample and the reference RNA sequence;

(iii) measuring the amounts of the amplified target viral RNA sequence and the amplified reference RNA sequence; and

(iv) determining the amount of the target viral RNA sequence present in the sample by comparing the amounts of the amplified target viral RNA sequence and the amplified reference RNA sequence.

Claim 57 (new): The process of claim 56, wherein the target viral RNA sequence is a human immunodeficiency virus (HIV) RNA sequence or a human cytomegalovirus (HCMV) RNA sequence.

Claim 58 (new): The process of claim 56, wherein a primer utilized in the polymerase chain reaction amplification includes a T-7 RNA polymerase binding sequence.

Claim 59 (new): The process of claim 56, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from a probe hybridized to the amplified target viral RNA sequence and (ii) the amount of signal obtained from a probe hybridized to the amplified reference RNA sequence.

Claim 60 (new): The process of claim 59, wherein the probes are labeled.

Claim 61 (new): The process of claim 60, wherein the label is an isotope or a fluorophore.

Claim 62 (new): The process of claim 59, wherein the reference RNA sequence comprises target viral RNA sequence and non-target viral RNA sequence.

Claim 63 (new): The process of claim 56, wherein the amount of the amplified target RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring

(i) the amount of signal obtained from the amplified target viral RNA sequence hybridized to a probe and (ii) the amount of signal obtained from the amplified reference RNA sequence hybridized to a probe.

Claim 64 (new): The process of claim 63, wherein primers utilized in the polymerase chain reaction amplification are labeled.

Claim 65 (new): The process of claim 64, wherein the label is an isotope or a fluorophore.

Claim 66 (new): The process of claim 63, wherein the reference RNA sequence comprises target viral RNA sequence and non-target viral RNA sequence.

Claim 67 (new): The process of claim 56, wherein the amount of the amplified target RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from the amplified target viral RNA sequence and (ii) the amount of signal obtained from the amplified reference RNA sequence.

Claim 68 (new): The process of claim 67, wherein primers utilized in the polymerase chain reaction amplification are labeled.

Claim 69 (new): The process of claim 68, wherein the label is an isotope or a fluorophore.

Claim 70 (new): The process of claim 67, wherein the reference RNA sequence comprises target viral RNA sequence and non-target viral RNA sequence.

Claim 71 (new): A process for quantitation of a target viral RNA sequence in a sample which comprises:

combining a predetermined quantity of a reference RNA sequence with the sample, wherein the reference RNA sequence is distinguishable from the target viral RNA sequence by size or by probes and wherein the reference RNA sequence is selected from the group consisting of (i) a reference RNA sequence which does not comprise the target viral RNA sequence, (ii) a reference RNA sequence which comprises the target viral RNA sequence and has substantially more nucleotides than the target viral RNA sequence, (iii) a reference RNA sequence which comprises the target viral RNA sequence with at least about 20 nucleotides less than the target viral RNA sequence and (iv) a reference RNA sequence which comprises target viral RNA sequence and non-target viral-RNA sequence.

simultaneously subjecting the target viral RNA sequence and the reference RNA sequence to polymerase chain reaction amplification under conditions appropriate to simultaneously amplify the target viral RNA sequence and the reference RNA sequence;

measuring the amounts of amplified target viral RNA sequence and amplified reference RNA sequence; and

determining the amount of the target viral RNA sequence present in the sample by comparing the amounts of the amplified target viral RNA sequence and the amplified reference RNA sequence.

Claim 72 (new): The process of claim 71, wherein the target viral RNA sequence is a human immunodeficiency virus (HIV) RNA sequence or a human cytomegalovirus (HCMV) RNA sequence.

Claim 73 (new): The process of claim 71, wherein a primer utilized in the polymerase chain reaction amplification includes a T-7 RNA polymerase binding sequence.

Claim 74 (new): The process of claim 71, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from a probe hybridized to the amplified target viral RNA sequence and (ii) the amount of signal obtained from a probe hybridized to the amplified reference RNA sequence.

Claim 75 (new): The process of claim 74, wherein the probes are labeled.

Claim 76 (new): The process of claim 75, wherein the label is an isotope or a fluorophore.

Claim 77 (new): The process of claim 71, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from the amplified target viral RNA sequence hybridized to a probe and (ii) the amount of signal obtained from the amplified reference RNA sequence hybridized to a probe.

Claim 78 (new): The process of claim 77, wherein primers utilized in the polymerase chain reaction amplification are labeled.

Claim 79 (new): The process of claim 78, wherein the label is an isotope or a fluorophore.

Claim 80 (new): The process of claim 71, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from the amplified target viral RNA sequence and (ii) the amount of signal obtained from the amplified reference RNA sequence.

Claim 81 (new): The process of claim 80, wherein primers utilized in the polymerase chain reaction amplification are labeled.

Claim 82 (new): The process of claim 81, wherein the label is an isotope or a fluorophore.

Claim 83 (new): A process for quantitation of a target viral RNA sequence in a sample which comprises:

combining a predetermined quantity of a reference RNA sequence with the sample, wherein the reference RNA sequence is distinguishable from the target viral RNA sequence by size or by probes and wherein the reference RNA sequence comprises target viral RNA sequence and non-target viral RNA sequence;

simultaneously subjecting the target viral RNA sequence and the reference RNA sequence to polymerase chain reaction amplification under conditions appropriate to simultaneously amplify the target viral RNA sequence and the reference RNA sequence;

measuring the amounts of the amplified target viral RNA sequence and the amplified reference RNA sequence; and

determining the amount of the target viral RNA sequence present in the sample by comparing the amounts of the amplified target viral RNA sequence and the amplified reference sequence.

Claim 84 (new): The process of claim 83, wherein the target viral RNA sequence is a human immunodeficiency virus (HIV) RNA segment or a human cytomegalovirus (HCMV) RNA sequence.

Claim 85 (new): The process of claim 83, wherein a primer utilized in the polymerase chain reaction amplification includes a T-7 RNA polymerase binding sequence.

Claim 86 (new): The process of claim 83, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from a probe hybridized to the amplified target viral RNA sequence and (ii) the amount of signal obtained from a probe hybridized to the amplified reference RNA sequence.

Claim 87 (new): The process of claim 86, wherein the hybridization probes are labeled.

Claim 88 (new): The process of claim 87, wherein the label is an isotope or a fluorophore.

Claim 89 (new): The process of claim 83, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from the amplified target viral RNA sequence hybridized to a probe and (ii) the amount of signal obtained from the amplified reference RNA sequence hybridized to a probe.

Claim 90 (new): The process of claim 89, wherein primers utilized in the polymerase chain reaction amplification are labeled.

Claim 91 (new): The process of claim 90, wherein the label is an isotope or a fluorophore.

Claim 92 (new): The process of claim 83, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from the amplified target viral RNA sequence and (ii) the amount of signal obtained from the amplified reference RNA sequence.

Claim 93 (new): The process of claim 92, wherein primers utilized in the polymerase chain reaction amplification are labeled.

Claim 94 (new): The process of claim 93, wherein the label is an isotope or a fluorophore.

Claim 95 (new): A process for quantitation of a target viral RNA sequence in a sample which comprises:

combining a predetermined quantity of a reference RNA sequence with the sample, wherein the reference RNA sequence is distinguishable from the target viral RNA sequence by size or by probes;

simultaneously subjecting the target viral RNA sequence and the reference RNA sequence to polymerase chain reaction amplification under conditions appropriate to simultaneously amplify the target viral RNA sequence and the reference RNA sequence;

measuring the amounts of amplified target viral RNA sequence and amplified reference RNA sequence; and

determining the amount of the target viral RNA sequence present in the sample by comparing the amounts of the amplified target viral RNA sequence and the amplified reference RNA sequence.

Claim 96 (new): The process of claim 95, wherein the target viral RNA sequence is a human immunodeficiency virus (HIV) RNA sequence or a human cytomegalovirus (HCMV) RNA sequence.

Claim 97 (new): The process of claim 95, wherein a primer utilized in the polymerase chain reaction amplification includes a T-7 RNA polymerase binding sequence.

Claim 98 (new): The process of claim 95, wherein the reference RNA sequence is selected from the group consisting of (i) a reference RNA sequence which does not comprise the target viral RNA sequence, (ii) a reference RNA sequence which comprises the target viral RNA sequence and has substantially more nucleotides than the target viral RNA sequence, (iii) a reference RNA sequence which comprises the target viral RNA sequence with at least about 20 nucleotides less than the target viral RNA sequence and (iv) a reference RNA sequence which comprises target viral RNA sequence and non-target viral-RNA sequence.

Claim 99 (new): The process of claim 98, wherein the reference RNA sequence does not comprise the target viral RNA sequence.

Claim 100 (new): The process of claim 98, wherein the reference RNA sequence comprises more nucleotides than the target viral RNA sequence.

Claim 101 (new): The process of claim 100, wherein the reference RNA sequence comprises the target viral RNA sequence with a multi-base insert.

Claim 102 (new): The process of claim 98, wherein the reference RNA sequence comprises fewer nucleotides than the target RNA sequence.

Claim 103 (new): The process of claim 102, wherein the reference RNA sequence comprises the target viral RNA sequence with a multi-base deletion.

Claim 104 (new): The process of claim 98, wherein the reference RNA sequence comprises target viral RNA sequence and non-target viral RNA sequence.

Claim 105 (new): The process of claim 95, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from a probe hybridized to the amplified target viral RNA sequence and (ii) the amount of signal obtained from a probe hybridized to the amplified reference RNA sequence.

Claim 106 (new): The process of claim 105, wherein the probes are labeled.

Claim 107 (new): The process of claim 106, wherein the label is an isotope or a fluorophore.

Claim 108 (new): The process of claim 95, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from the amplified target viral RNA sequence hybridized to a probe and (ii) the amount of signal obtained from the amplified reference RNA sequence hybridized to a probe.

Claim 109 (new): The process of claim 108, wherein primers utilized in the polymerase chain reaction amplification are labeled.

Claim 110 (new): The process of claim 109, wherein the label is an isotope or a fluorophore.

Claim 111 (new): The process of claim 95, wherein the amount of the amplified target viral RNA sequence and the amount of the amplified reference RNA sequence are measured by measuring (i) the amount of signal obtained from the amplified target viral RNA sequence and (ii) the amount of signal obtained from the amplified reference RNA sequence.

Claim 112 (new): The process of claim 111, wherein primers utilized in the polymerase chain reaction amplification are labeled.

Claim 113 (new): The process of claim 112, wherein the label is an isotope or a fluorophore.